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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
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10/840,182

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Michael D. Cleary

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10/18/2006

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EXAMINER

PANDE, SUCHIRA

ART UNIT

PAPER NUMBER

1637

DATE MAILED: 10/18/2006

Please find below and/or attached an Office communication concerning this application or proceeding.

**Office Action Summary**

Application No.

10/840,182

Applicant(s)

CLEARY ET AL.

Examiner

Suchira Pande

Art Unit

1637

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --  
**Period for Reply**

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

**Status**

- 1) ☒ Responsive to communication(s) filed on 30 August 2006.
- 2a) ☐ This action is **FINAL**. 2b) ☒ This action is non-final.
- 3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

**Disposition of Claims**

- 4) ☒ Claim(s) 1-37 is/are pending in the application.
- 4a) Of the above claim(s) 8,9,12,14-17,20-22,24-27,29,30 and 32 is/are withdrawn from consideration.
- 5) ☐ Claim(s) \_\_\_\_\_ is/are allowed.
- 6) ☒ Claim(s) 1-7,10,11,13,18,19,23,28,31 and 33-37 is/are rejected.
- 7) ☐ Claim(s) \_\_\_\_\_ is/are objected to.
- 8) ☐ Claim(s) \_\_\_\_\_ are subject to restriction and/or election requirement.

**Application Papers**

- 9) ☒ The specification is objected to by the Examiner.
- 10) ☒ The drawing(s) filed on 05/05/2004 is/are: a) ☐ accepted or b) ☒ objected to by the Examiner.  
Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).  
Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).
- 11) ☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

**Priority under 35 U.S.C. § 119**

- 12) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).  
a) ☐ All b) ☐ Some \* c) ☐ None of:
1. ☐ Certified copies of the priority documents have been received.
  2. ☐ Certified copies of the priority documents have been received in Application No. \_\_\_\_\_.
  3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

\* See the attached detailed Office action for a list of the certified copies not received.

**Attachment(s)**

- |  |   |
|--|---|
| 1) <input checked="" type="checkbox"/> Notice of References Cited (PTO-892)  | 4) <input type="checkbox"/> Interview Summary (PTO-413)<br>Paper No(s)/Mail Date. _____ |
| 2) <input type="checkbox"/> Notice of Draftsperson's Patent Drawing Review (PTO-948)   | 5) <input type="checkbox"/> Notice of Informal Patent Application                       |
| 3) <input checked="" type="checkbox"/> Information Disclosure Statement(s) (PTO/SB/08)<br>Paper No(s)/Mail Date <u>11/18/05; 12/6/04</u> | 6) <input type="checkbox"/> Other: _____  |

## **DETAILED ACTION**

### ***Election/Restrictions***

1. Applicant's election with traverse of Group I claims (1-37) in the reply filed on August 30, 2006 is acknowledged. Applicant further elected the species nitrogenous base for purine or pyrimidine analog; a small molecule binding partner for species of tags; RNA amplification for species of methods for processing separated RNA; constitutively active promoter for species of promoters; and a replicable vector for species of vector.

Consequently, claims 8-9, 12, 14-17, 20-22, 24-27, 29-30 and 32 are withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to a nonelected claims, there being no allowable generic or linking claim.

Claims 1-7, 10-11, 13, 18-19, 23, 28, 31 and 33-37 are under examination in this application.

2. Applicant is reminded that upon the cancellation of claims to a non-elected invention, the inventorship must be amended in compliance with 37 CFR 1.48(b) if one or more of the currently named inventors is no longer an inventor of at least one claim remaining in the application. Any amendment of inventorship must be accompanied by a request under 37 CFR 1.48(b) and by the fee required under 37 CFR 1.17(i).

### ***Drawings***

3. New corrected drawings in compliance with 37 CFR 1.121(d) are required in this application because Figure 3 is in black and white and does not show the colors red and green representing the induced and repressed genes as described in the brief

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description of drawings. Applicant is advised to provide colored drawing for Figure 3.

The corrected drawings are required in reply to the Office action to avoid abandonment of the application. The requirement for corrected drawings will not be held in abeyance.

### ***Specification***

4. The disclosure is objected to because of the following informalities: The use of trademark FLAG has been noted in this application on page 13. Numerous dyes names such as Cy3, Cy5, Texas Red etc. that are trademarks appear on pages 17 and several following pages. Trademark should be capitalized wherever it appears followed by superscript TM and be accompanied by the generic terminology.

Although the use of trademarks is permissible in patent applications, the proprietary nature of the marks should be respected and every effort made to prevent their use in any manner which might adversely affect their validity as trademarks.

Applicant is advised to scan the application for all the places where trademarks appear and ensure they are appropriately used in the disclosure.

Appropriate correction is required.

### ***Claim Rejections - 35 USC § 103***

5. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

(a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negated by the manner in which the invention was made.

6. This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of

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the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

7. Claims 1-7, 10-11, 13, 18-19, 23-28 and 31 are rejected under 35 U.S.C. 103(a) as being unpatentable over Trudeau et. al. (2001) Human Gene Therapy 12:1673-1680 in view of Johnson et. al. (1991) Proc. Natl. Acad. Sci. USA Vol. 88: pp. 5287-5291 and Rana P.G. Pub 2004/0175732 filed on November 17, 2003 with a priority date of November 15, 2002.

A. Regarding claim 1, Trudeau et. al. teaches: *A method of biosynthetically labeling RNA in a cell of interest, the method comprising:*

*contacting said cell with a purine or pyrimidine analog having a reactive moiety not normally present in RNA, (see page 1676 par. 4 where purine analog allopurinol and page 1679 par. 2 where purine analog thioguanine (6-TG) are taught. 6-TG contains a thio reactive moiety not normally present in RNA. See page 1675 par. 2 and 4 where contacting said cell with purine analog allopurinol and 6-TG are taught)*

*wherein said cell comprises a phosphoribosyltransferase or nucleoside kinase or phosphorylase that can specifically incorporate said purine or pyrimidine analog into the corresponding nucleotide, and wherein said purine or pyrimidine analog is incorporated into RNA synthesized by said cell; (see page 1676 par. 4 where hypoxanthine-guanine*

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phosphoribosyltransferase (HGPRT) enzyme is taught. HGPRT from *Trypanosoma brucei* (TbHGPRT) is capable of converting allopurinol to its ribonucleoside 5'-monophosphate (HPPR-MP). HPPR-MP is then converted to 4-aminopyrazolo pyrimidine ribonucleotide (APPR-MP) via the adenylosuccinate synthetase and the adenylosuccinate lyase enzymes. This APPR-MP is metabolized into triphosphates and incorporated into the parasite RNA. (see page 1676 par. 4).

See page 1679, par. 2 where conversion of 6-TG to its nucleotide form by human HGPRT is taught.

See Fig. 4 on page 1679, where uptake of  $^3\text{H}$  allopurinol by cells containing TbHGPRT is taught.

*obtaining RNA from said cell;* (see page 1676 where Trudeau et. al. teach metabolism of APPR-MP into nucleotides and its incorporation into parasite RNA, leading to inhibition of the RNA replication chain but does not actually teach obtaining RNA from said cell).

Regarding claim 2, Trudeau et. al. teaches *wherein sequences encoding said phosphoribosyltransferase or nucleoside kinase are operably linked to a promoter that is active or can be activated in said cell.* (see page 1674 par. 3 where construction of pTbIGFP construct by inserting HGPRT gene into the multiple cloning site of AP2, a retroviral vector expressing enhanced green fluorescent protein (EGFP) is taught. Here sequence encoding HGPRT gene is operably linked to a promoter that drives the expression of EGFP in this retroviral vector.)

Regarding claim 3, Trudeau et. al. teaches *wherein said sequences encoding said phosphoribosyltransferase or nucleoside kinase are exogenous to the cell of interest*. (see page 1675 par. 4 where Trudeau et. al. teaches uptake of allopurinol by stably transduced cell lines A549, H322 and DA3. Here HGPRT sequences are exogenous (derived from *Trypanosoma brucei*) to the cells (A549, H322 and DA3) of interest).

Regarding claim 4, Trudeau et. al. teaches *wherein said reactive moiety is at least one thiol group*. (see page 1679, par. 2 where purine analog 6-thioguanine is taught).

Regarding claim 23, Trudeau et. al. teaches *wherein said promoter is constitutively active in said cell of interest*. (see page 1676 Fig. 1 and Fig. Legend for Fig. 1 A, where plasmid pTbIGFP expresses HGPRT gene under control of a constitutively active CMV, cytomegalovirus promoter element. Thus Trudeau et. al. teaches wherein said promoter is constitutively active in said cell of interest).

Regarding claim 28, Trudeau et. al. teaches *wherein said sequences encoding said phosphoribosyltransferase or nucleoside kinase are introduced into said cell of interest on a replicable vector*. (see page 1674 par. 6 where E.coli cells containing TbHGPRT gene was overexpressed, here TbHGPRT is a cDNA clone provided by B. Ullman. This TbHGPRT cDNA clone is obviously present on a replicable vector that has an inducible promoter capable of expression in *E.coli* cell. Thus Trudeau et. al. teaches wherein said sequences encoding said phosphoribosyltransferase are introduced into said cell of interest on a replicable vector).

Regarding claim 31, Trudeau et. al. teaches *wherein said purine or pyrimidine analog is provided in the form of a nitrogenous base*. (see page 1676 par. 4 where purine analog allopurinol an isomer of hypoxanthine a natural base is taught. Thus Trudeau et. al. teaches wherein said purine or pyrimidine analog is provided in the form of a nitrogenous base (allopurinol).

Regarding claim 1, Trudeau et. al. does not teach :

*obtaining RNA from said cell; and conjugating a tag to said reactive moiety.*

B) Regarding claim 1, Johnson et. al. (1991) teach: Biosynthetically labeling newly synthesized RNA with thiol moiety to generate thiolated RNA (see page 5287 par. 2). They teach use of two thiol precursors 6- thioguanosine and 4- thiouridine to label RNA. Only 4-thiouridine was used for labeling RNA as no binding to mercurated agarose affinity matrix was observed when 6- thioguanosine was used as a precursor (see page 5287 par. 4.). Thiol labeled RNA formed after using 4- thiouridine as a precursor was bound to Affi-Gel 501 to obtain labeled RNA from cell. (see page 5288 par. 1). Thus Johnson et. al. teach *obtaining thiol labeled RNA from said cell;*

Regarding claims 4, Johnson et. al. teaches *wherein said reactive moiety is at least one thiol group*. (see page 5287 par. 4 where 4-thiouridine containing at least one thiol group is taught)

Regarding claims 5, Johnson et. al. teaches *wherein said purine or pyrimidine analog is a uracil analog* (see page 5287 par. 4 where 4-thiouridine an analog of uracil is taught).

Johnson et. al. do not teach: *conjugating a tag to said reactive moiety.*

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C) Regarding claim 1, Rana teaches: *conjugating a tag to said reactive moiety*. (see page 1 par. 0004 where Rana teaches addition of tags such as biotin, psoralen-biotin and 4-thiobiotin to isolated RNA. See Fig. 8 D and page 6, par. 0065 where Rana teaches biotinylated RNA molecule labeled with 4-thio uridine or 6-thioguanosine. Binding to streptavidin coated beads selectively enriches RNA isolated from cell for RNA molecules containing biotin tag.

Regarding claim 6, Rana teaches wherein *said tag is a small molecule binding partner*. (see page 1, par. 0004 where small molecule binding partner biotin is taught as a tag)

Regarding claim 7, Rana teaches *wherein said tag is biotin*. (see page 1, par. 0004 where small molecule binding partner biotin is taught as a tag)

Regarding claim 10, Rana teaches *method further comprising the step of binding a specific binding partner to said tag*. (see page 6, par. 0065 where the step of binding streptavidin, a specific binding partner to said tag biotin is taught).

Regarding claim 11, Rana teaches *wherein said specific binding partner is conjugated to an insoluble substrate for affinity chromatography, and wherein said biosynthetically labeled RNA is separated from non-labeled RNA*. (see page 6, par. 0065 where use of streptavidin coated beads to separate biotin labeled RNA from total RNA is taught. Here specific binding partner (streptavidin) is conjugated to an insoluble substrate (beads) for affinity chromatography, and wherein said biosynthetically labeled RNA is separated from non-labeled RNA.

Regarding claim 13, Rana teaches *wherein said separated RNA is amplified*.  
(see page 6, par. 0069 where amplification of RNA by reverse transcription is taught).

Regarding claim 18, Rana teaches *wherein said specific binding partner is conjugated to a detectable label*. (see page 13, par. 0145 where avidin and streptavidin conjugated to different detectable labels magnetic particles, superparamagnetic microspheres are taught. Further Rana teaches custom synthesis of beads when biotin/avidin or biotin/streptavidin system is used. Thereby Rana teaches the specific binding partner could be labeled with any other desired detectable label )

Regarding claim 19, Rana teaches *wherein said detectable label is a fluorochrome, radiolabel, heavy metal label, or enzyme conjugate*. (see page 13, par. 0141 where detectable labels such as flurochrome such as fluorescein, radiolabel such as <sup>32</sup>P etc are taught)

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to combine the teachings of Johnson et. al. and Rana in the method of labeling RNA taught by Trudeau et. al.

Trudeau et. al. teach the entire method and explain the principle in detail how parasite encoded phosphoribosyltransferase enzyme can be used to incorporate purine analogs into newly synthesized RNA but does not actually enunciate the method how this thio labeled RNA can be isolated from the cell. Johnson et. al. provide the details how thio labeled RNA can be obtained free of unlabeled RNA. To one of ordinary skill in the art it would be evident that there was reasonable expectation of success that they

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would be able to isolate thiolated RNA by practicing the method of Johnson et. al. in the cells that had been labeled using the method of Trudeau et. al.

The motivation to combine the method of Rana in the method of Trudeau et. al. and Johnson et. al. is provided by both Johnson et. al. and Rana. Johnson et. al. point out the drawback associated with the Affi-Gel 501 binding and recovery of 4-thiouridine labeled RNA. (see Johnson et. al. page 5288 par. 1 where they teach "These conditions allowed 47% recovery of labeled RNA" thus 53% of the thiol labeled newly synthesized RNA is lost during washes after binding of RNA to the affinity matrix. Johnson et. al. go on to say " Binding of RNA to the affinity matrix was done at 4°C in the dark, since 4-thiouridine is photoactivatable". Rana teaches conjugating a tag such as biotin to the thiolated RNA. The conjugation to biotin for example now allows one of ordinary skill in the art to exploit the biotin/streptavidin chemistry to isolate thiolated labeled RNA very efficiently.

8. Claims 33-35 and 37 are rejected under 35 U.S.C. 103(a) as being unpatentable over Al-Anouti et. al. (January 2003) Biochemical and Biophysical Research Communications vol. 302: pp. 316-323 in view of Johnson et. al. (1991) Proc. Natl. Acad. Sci. USA Vol. 88: pp. 5287-5291

A) Regarding claim 33, Al-Anouti et. al. teach: *A method of biosynthetically labeling RNA in a cell of interest, the method comprising:*

*contacting said cell with a uracil analog* (see page 316 par. 1 where contacting the cells with uracil analog 5-fluoro-2'-deoxyuridine (FUDR) is taught)

*having a reactive thiol moiety not normally present in RNA,*

*wherein said cell comprises a uracil phosphoribosyltransferase (UPRT) that can convert said uracil analog to the corresponding uridine monophosphate; (see page 316 par. 1 where a uracil phosphoribosyltransferase (UPRT) from Toxoplasma gondii (T. gondii) that can convert said uracil analog to the corresponding uridine monophosphate 5-fluorodeoxyuridine monophosphate is taught);*

*wherein said uridine analog is incorporated into RNA synthesized by said cell.*

(The uridine monophosphate unit thus generated by UPRT would be incorporated by T. gondii because UPRT is the only operative enzyme of the pyrimidine salvage pathway that uses preformed pyrimidines at the nucleotide (see page 316 par. 1). However 5-fluorodeoxyuridine monophosphate is toxic to the cell because it inhibits synthesis of thymidine monophosphate and hence FUDR is used to select T. gondii where UPRT expression is halted. Consequently, Al-Anouti et. al. does not show the incorporation of said uridine analog into RNA synthesized by said cell.

Regarding claim 34, Al-Anouti et. al. teaches *wherein sequences encoding said UPRT are operably linked to a promoter that is active or can be activated in said cell.* (see page 317 par. 4 where no of plasmids encoding UPRT under control of modified promoter of T. gondii surface antigen 1(SAG1) and their derivatives are taught. The plasmids containing UPRT gene under control of T.gondii promoter and pUC19UPRT plasmids all contain UPRT operably linked to a promoter that is active or can be activated in appropriate Human Foreskin Fibroblasts or E.coli cells respectively.)

Regarding claim 35, Al-Anouti et. al. teaches *wherein said sequences encoding said UPRT are exogenous to the cell of interest.* (see page 317, par. 4 where pUC19UPRT plasmid is taught.. These pUC based plasmids can be propagated in *E.coli*. Thus Al-Anouti et. al. teaches UPRT gene from *T.gondii* is exogenous to the cell of interest.)

Regarding claim 37, Al-Anouti et. al. teaches *wherein said UPRT is Toxoplasma gondii UPRT or a functional derivative thereof.* (see page 316 title and abstract where *Toxoplasma gondii* uracil phosphoribosyltransferase (TgUPRT) is taught).

Regarding claim 33, Al-Anouti et. al. does not teach:

*a uracil analog having a reactive thiol moiety not normally present in RNA, wherein said uridine analog is incorporated into RNA synthesized by said cell.*

B) Regarding claim 33, Johnson et. al. teach:

*a uracil analog having a reactive thiol moiety not normally present in RNA,* (see page 5287 par. 2 where they teach biosynthetically labeling newly synthesized RNA with thiol moiety to generate thiolated RNA. They teach use of two compounds 6-thioguanosine and 4- thiouridine both uracil analogs have a reactive thiol moiety not normally present in RNA..

*wherein said uridine analog is incorporated into RNA synthesized by said cell.* (see page 5287 par. 4. where Johnson et. al. teaches 4-thiouridine was used for labeling RNA. Thiol labeled RNA formed after using 4- thiouridine as a precursor was bound to Affi-Gel 501 to obtain labeled RNA from cell. (see page 5288 par. 1). Thus

Johnson et. al. teaches wherein said uridine analog is incorporated into RNA synthesized by said cell;

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to combine the teachings of Johnson et. al. in the method of Al-Anouti et. al.

Al-Anouti et. al. teach the entire method and explain the principle in detail how parasite encoded UPRT enzyme can be used to incorporate uridine analogs into newly synthesized RNA but did not use thiol containing analogs as they were interested in isolating mutants of UPRT and therefore also did not show incorporation of analog used into newly synthesized RNA.

The motivation to practice the method of Johnson et. al. in the method of Al-Anouti et. al. is provided by Johnson et. al. who state "In this report we show the analysis of thiolated, newly synthesized RNA, in addition to yielding data on relative transcription rates for specific transcripts under -----, can provide estimates of transcript half-life, which are not subject to the same limitations as studies employing transcriptional inhibitors or pulse-chase methods." (see page 5287 par. 2).

Thus by combining the two methods one of ordinary skill in the art has reasonable expectation of success in being able to thiolate newly synthesized RNA from cells containing UPRT enzyme.

9. Claim 36 is rejected under 35 U.S.C. 103(a) as being unpatentable over Al-Anouti et. al. (January 2003) Biochemical and Biophysical Research Communications

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vol. 302: pp. 316-323 and Johnson et. al. (1991) Proc. Natl. Acad. Sci. USA Vol. 88: pp. 5287-5291 as applied to claims 33-35 and 37 above further in view of Iltzsch and Tankersley (1994) Biochem Pharm. Vol. 48 (4): 781-792 cited by applicant in IDS.

Regarding claim 36, Al-Anouti et. al. and Johnson et. al. teaches the method of claim 33 but do not teach uracil analog 2,4 dithiouracil.

Regarding claim 36, Iltzsch and Tankersley teach uracil analog 2,4 dithiouracil. (see page 781 abstract where 2,4-dithiouracil a uracil analog containing thiol moiety not normally present in cell is taught as a substrate for *T. gondii* enzyme UPRT).

It would have been obvious to one of ordinary skill in the art to use uracil analog 2,4 dithiouracil taught by Iltzsch and Tankersley in the method of Al-Anouti et. al. & Johnson et. al. The motivation to do so is provided by Johnson et. al. who point out the drawback associated with use of 4-thiouridine labeled RNA. (see Johnson et. al. page 5288 par. 1 where they state " Binding of RNA to the affinity matrix was done at 4°C in the dark, since 4-thiouridine is photoactivatable". Thus by using 2,4 dithiouracil to thiolate the newly synthesized RNA one of ordinary skill in the art would have access to thiol labeled RNA that was not sensitive to light hence the studies could be conducted normally in presence of light.

### **Conclusion**

All claims under consideration 1-7,10-11, 13, 18-19, 23, 28, 31, 33-37 are rejected.

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Any inquiry concerning this communication or earlier communications from the examiner should be directed to Suchira Pande whose telephone number is 571-272-9052. The examiner can normally be reached on 8:30 am -5:00 pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 571-272-0782. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov>. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

Suchira Pande  
Examiner  
Art Unit 1637



JEFFREY FREDMAN  
PRIMARY EXAMINER

10/12/01